



NREL Seq listing 399697
SEQUENCE LISTING

<110> NATIONAL RENEWABLE ENERGY LABORATORY
Himmel, Michael E.
Adney, William

<120> ENDOGLUCANASE MUTANTS AND MUTANT HYDROLYTIC DEPOLYMERIZING
ENZYMES AND USES THEREOF

<130> NREL 99-38

<140> US 09/997,504

<141> 2001-11-19

<150> US 60/134,925

<151> 1999-05-19

<160> 14

<170> PatentIn version 3.2

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<212> DNA

<213> Acidothermus cellulolyticus

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<211> 38

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<213> Acidothermus cellulolyticus

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<221> misc_feature

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<213> Acidothermus cellulolyticus

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<212> DNA
<213> Acidothermus cellulolyticus

<220>
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<212> DNA
<213> Acidothermus cellulolyticus

<220>
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ctctggctac gcgactaccg cagcatgctc gaccagataa agtcgctcgg ctacaacaca 180
atccggctgc cgtactctga cgacattctc aagccgggca ccatgccgaa cagcatcaat 240
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<210> 8
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 <212> PRT
 <213> Acidothermus cellulolyticus

<220>
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<400> 8

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Asn Asn Val Pro Val Arg Ile Ala Gly Ile Asn Trp Phe Gly Phe Glu
 20 25 30

Thr Cys Asn Tyr Val Val His Gly Leu Trp Ser Arg Asp Tyr Arg Ser
 35 40 45

Met Leu Asp Gln Ile Lys Ser Leu Gly Tyr Asn Thr Ile Arg Leu Pro
 50 55 60

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Tyr Ser Asp Asp Ile Leu Lys Pro Gly Thr Met Pro Asn Ser Ile Asn
65 70 75 80

Phe Tyr Gln Met Asn Gln Asp Leu Gln Gly Leu Thr Ser Leu Gln Val
85 90 95

Met Asp Lys Ile Val Ala Tyr Ala Gly Gln Ile Gly Leu Arg Ile Ile
100 105 110

Leu Asp Arg His Arg Pro Asp Cys Ser Gly Gln Ser Ala Leu Trp Tyr
115 120 125

Thr Ser Ser Val Ser Glu Ala Thr Trp Ile Ser Asp Leu Gln Ala Leu
130 135 140

Ala Gln Arg Tyr Lys Gly Asn Pro Thr Val Val Gly Phe Asp Leu His
145 150 155 160

Asn Glu Pro His Asp Pro Ala Cys Trp Gly Cys Gly Asp Pro Ser Ile
165 170 175

Asp Trp Arg Leu Ala Ala Glu Arg Ala Gly Asn Ala Val Leu Ser Val
180 185 190

Asn Pro Asn Leu Leu Ile Phe Val Glu Gly Val Gln Ser Tyr Asn Gly
195 200 205

Asp Ser Tyr Trp Trp Gly Gly Asn Leu Gln Gly Ala Gly Gln Tyr Pro
210 215 220

Val Val Leu Asn Val Pro Asn Arg Leu Val Tyr Ser Ala His Asp Tyr
225 230 235 240

Ala Thr Ser Val Tyr Pro Gln Thr Trp Phe Ser Asp Pro Thr Phe Pro
245 250 255

Asn Asn Met Pro Gly Ile Trp Asn Lys Asn Trp Gly Tyr Leu Phe Asn
260 265 270

Gln Asn Ile Ala Pro Val Trp Leu Gly Glu Phe Gly Thr Thr Leu Gln
275 280 285

Ser Thr Thr Asp Gln Thr Trp Leu Lys Thr Leu Val Gln Tyr Leu Arg
290 295 300

Pro Thr Ala Gln Tyr Gly Ala Asp Ser Phe Gln Trp Thr Phe Trp Ser
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305 310 315 320

Trp Asn Pro Asp Ser Gly Asp Thr Gly Gly Ile Leu Lys Asp Asp Trp
325 330 335

Gln Thr Val Asp Thr Val Lys Asp Gly Tyr Leu Ala Pro Ile Lys Ser
340 345 350

Ser Ile Phe Asp Pro Val Gly
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<210> 9
<211> 1303
<212> DNA
<213> Acidothermus cellulolyticus

<220>
<221> misc_feature
<223> DNA sequence for Y245G Mutant SEQ ID: 9

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ctctggtcac gcgactaccg cagcatgctc gaccagataa agtcgctcgg ctacaacaca 180
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ttttaccaga tgaatcagga cctgcagggt ctgacgtcct tgcaggatcat ggacaaaatc 300
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agcgggcagt cggcgctgtg gtacacgagc agcgtctcgg aggctacgtg gatttccgac 420
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aacgagccgc atgacccggc ctgctggggc tgcggcgatc cgagcatcga ctggcgattg 540
gccgccgagc gggccggaaa cgccgtgctc tcggtgaatc cgaacctgct cattttcgtc 600
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ccgctgcacc gcgagttacc aggtcaacag cgattggggc aat 1303

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<211> 358
<212> PRT
<213> Acidothermus cellulolyticus

<220>
<221> misc_feature
<223> Translated amino acid sequence for Y245G mutant SEQ ID:10

<400> 10

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1 5 10 15

Asn Asn Val Pro Val Arg Ile Ala Gly Ile Asn Trp Phe Gly Phe Glu
20 25 30

Thr Cys Asn Tyr Val Val His Gly Leu Trp Ser Arg Asp Tyr Arg Ser
35 40 45

Met Leu Asp Gln Ile Lys Ser Leu Gly Tyr Asn Thr Ile Arg Leu Pro
50 55 60

Tyr Ser Asp Asp Ile Leu Lys Pro Gly Thr Met Pro Asn Ser Ile Asn
65 70 75 80

Phe Tyr Gln Met Asn Gln Asp Leu Gln Gly Leu Thr Ser Leu Gln Val
85 90 95

Met Asp Lys Ile Val Ala Tyr Ala Gly Gln Ile Gly Leu Arg Ile Ile
100 105 110

Leu Asp Arg His Arg Pro Asp Cys Ser Gly Gln Ser Ala Leu Trp Tyr
115 120 125

Thr Ser Ser Val Ser Glu Ala Thr Trp Ile Ser Asp Leu Gln Ala Leu
130 135 140

Ala Gln Arg Tyr Lys Gly Asn Pro Thr Val Val Gly Phe Asp Leu His
145 150 155 160

Asn Glu Pro His Asp Pro Ala Cys Trp Gly Cys Gly Asp Pro Ser Ile
165 170 175

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Asp Trp Arg Leu Ala Ala Glu Arg Ala Gly Asn Ala Val Leu Ser Val
180 185 190

Asn Pro Asn Leu Leu Ile Phe Val Glu Gly Val Gln Ser Tyr Asn Gly
195 200 205

Asp Ser Tyr Trp Trp Gly Gly Asn Leu Gln Gly Ala Gly Gln Tyr Pro
210 215 220

Val Val Leu Asn Val Pro Asn Arg Leu Val Tyr Ser Ala His Asp Tyr
225 230 235 240

Ala Thr Ser Val Gly Pro Gln Thr Trp Phe Ser Asp Pro Thr Phe Pro
245 250 255

Asn Asn Met Pro Gly Ile Trp Asn Lys Asn Trp Gly Tyr Leu Phe Asn
260 265 270

Gln Asn Ile Ala Pro Val Trp Leu Gly Glu Phe Gly Thr Thr Leu Gln
275 280 285

Ser Thr Thr Asp Gln Thr Trp Leu Lys Thr Leu Val Gln Tyr Leu Arg
290 295 300

Pro Thr Ala Gln Tyr Gly Ala Asp Ser Phe Gln Trp Thr Phe Trp Ser
305 310 315 320

Trp Asn Pro Asp Ser Gly Asp Thr Gly Gly Ile Leu Lys Asp Asp Trp
325 330 335

Gln Thr Val Asp Thr Val Lys Asp Gly Tyr Leu Ala Pro Ile Lys Ser
340 345 350

Ser Ile Phe Asp Pro Val
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<210> 11
<211> 1337
<212> DNA
<213> Acidothermus cellulolyticus

<220>
<221> misc_feature
<223> DNA sequence for W42R Mutant SEQ ID: 11

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ttttaccaga tgaatcagga cctgcagggt ctgacgtcct tgcagggtcat ggacaaaatc 300
gtcgcgtacg ccggtcagat cggcctgcgc atcattcttg accgccaccg accggattgc 360
agcgggcagt cggcgctgtg gtacacgagc agcgtctcgg aggctacgtg gatttccgac 420
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aacgagccgc atgaccgggc ctgctggggc tgcggcgatc cgagcatcga ctggcgattg 540
gccgccgagc gggccggaaa cgccgtgctc tcggtgaatc cgaacctgct cattttcgtc 600
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ggccgtgaca aattccg 1337

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<210> 12
 <211> 358
 <212> PRT
 <213> Acidothermus cellulolyticus

<220>
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 <223> Translated amino acid sequence for W42R mutant SEQ ID: 12
 <400> 12

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Asn Asn Val Pro Val Arg Ile Ala Gly Ile Asn Trp Phe Gly Phe Glu
 20 25 30

NREL Seq listing 399697

Thr Cys Asn Tyr Val Val His Gly Leu Arg Ser Arg Asp Tyr Arg Ser
35 40 45

Met Leu Asp Gln Ile Lys Ser Leu Gly Tyr Asn Thr Ile Arg Leu Pro
50 55 60

Tyr Ser Asp Asp Ile Leu Lys Pro Gly Thr Met Pro Asn Ser Ile Asn
65 70 75 80

Phe Tyr Gln Met Asn Gln Asp Leu Gln Gly Leu Thr Ser Leu Gln Val
85 90 95

Met Asp Lys Ile Val Ala Tyr Ala Gly Gln Ile Gly Leu Arg Ile Ile
100 105 110

Leu Asp Arg His Arg Pro Asp Cys Ser Gly Gln Ser Ala Leu Trp Tyr
115 120 125

Thr Ser Ser Val Ser Glu Ala Thr Trp Ile Ser Asp Leu Gln Ala Leu
130 135 140

Ala Gln Arg Tyr Lys Gly Asn Pro Thr Val Val Gly Phe Asp Leu His
145 150 155 160

Asn Glu Pro His Asp Pro Ala Cys Trp Gly Cys Gly Asp Pro Ser Ile
165 170 175

Asp Trp Arg Leu Ala Ala Glu Arg Ala Gly Asn Ala Val Leu Ser Val
180 185 190

Asn Pro Asn Leu Leu Ile Phe Val Glu Gly Val Gln Ser Tyr Asn Gly
195 200 205

Asp Ser Tyr Trp Trp Gly Gly Asn Leu Gln Gly Ala Gly Gln Tyr Pro
210 215 220

Val Val Leu Asn Val Pro Asn Arg Leu Val Tyr Ser Ala His Asp Tyr
225 230 235 240

Ala Thr Ser Val Tyr Pro Gln Thr Trp Phe Ser Asp Pro Thr Phe Pro
245 250 255

Asn Asn Met Pro Gly Ile Trp Asn Lys Asn Trp Gly Tyr Leu Phe Asn
260 265 270

Gln Asn Ile Ala Pro Val Trp Leu Gly Glu Phe Gly Thr Thr Leu Gln
275 280 285

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Ser Thr Thr Asp Gln Thr Trp Leu Lys Thr Leu Val Gln Tyr Leu Arg
290 295 300

Pro Thr Ala Gln Tyr Gly Ala Asp Ser Phe Gln Trp Thr Phe Trp Ser
305 310 315 320

Trp Asn Pro Asp Ser Gly Asp Thr Gly Gly Ile Leu Lys Asp Asp Trp
325 330 335

Gln Thr Val Asp Thr Val Lys Asp Gly Tyr Leu Ala Pro Ile Lys Ser
340 345 350

Ser Ile Phe Asp Pro Val
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<210> 13
<211> 1337
<212> DNA
<213> Acidothermus cellulolyticus

<220>
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<223> DNA sequence for Y82R Mutant SEQ ID: 13

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 <212> PRT
 <213> Acidothermus cellulolyticus

<220>
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 <223> Translated amino acid sequence for Y82R mutant SEQ ID: 14
 <400> 14

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Asn Asn Val Pro Val Arg Ile Ala Gly Ile Asn Trp Phe Gly Phe Glu
 20 25 30

Thr Cys Asn Tyr Val Val His Gly Leu Trp Ser Arg Asp Tyr Arg Ser
 35 40 45

Met Leu Asp Gln Ile Lys Ser Leu Gly Tyr Asn Thr Ile Arg Leu Pro
 50 55 60

Tyr Ser Asp Asp Ile Leu Lys Pro Gly Thr Met Pro Asn Ser Ile Asn
 65 70 75 80

Phe Arg Gln Met Asn Gln Asp Leu Gln Gly Leu Thr Ser Leu Gln Val
 85 90 95

Met Asp Lys Ile Val Ala Tyr Ala Gly Gln Ile Gly Leu Arg Ile Ile
 100 105 110

Leu Asp Arg His Arg Pro Asp Cys Ser Gly Gln Ser Ala Leu Trp Tyr
 115 120 125

Thr Ser Ser Val Ser Glu Ala Thr Trp Ile Ser Asp Leu Gln Ala Leu
 130 135 140

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Ala Gln Arg Tyr Lys Gly Asn Pro Thr Val Val Gly Phe Asp Leu His
145 150 155 160

Asn Glu Pro His Asp Pro Ala Cys Trp Gly Cys Gly Asp Pro Ser Ile
165 170 175

Asp Trp Arg Leu Ala Ala Glu Arg Ala Gly Asn Ala Val Leu Ser Val
180 185 190

Asn Pro Asn Leu Leu Ile Phe Val Glu Gly Val Gln Ser Tyr Asn Gly
195 200 205

Asp Ser Tyr Trp Trp Gly Gly Asn Leu Gln Gly Ala Gly Gln Tyr Pro
210 215 220

Val Val Leu Asn Val Pro Asn Arg Leu Val Tyr Ser Ala His Asp Tyr
225 230 235 240

Ala Thr Ser Val Tyr Pro Gln Thr Trp Phe Ser Asp Pro Thr Phe Pro
245 250 255

Asn Asn Met Pro Gly Ile Trp Asn Lys Asn Trp Gly Tyr Leu Phe Asn
260 265 270

Gln Asn Ile Ala Pro Val Trp Leu Gly Glu Phe Gly Thr Thr Leu Gln
275 280 285

Ser Thr Thr Asp Gln Thr Trp Leu Lys Thr Leu Val Gln Tyr Leu Arg
290 295 300

Pro Thr Ala Gln Tyr Gly Ala Asp Ser Phe Gln Trp Thr Phe Trp Ser
305 310 315 320

Trp Asn Pro Asp Ser Gly Asp Thr Gly Gly Ile Leu Lys Asp Asp Trp
325 330 335

Gln Thr Val Asp Thr Val Lys Asp Gly Tyr Leu Ala Pro Ile Lys Ser
340 345 350

Ser Ile Phe Asp Pro Val
355